(FILE 'HOME' ENTERED AT 18:02 14 ON 05 NOV 2001)

FILE 'MEDLINE, AGRICOLA, CAPLUS, BIOSIS, EMBASE, WPIDS' ENTERED AT
18:02:22 ON 05 NOV 2001
L1 1 S (MANC OR MANB) AND (RFBK OR RFBM)

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L1 ANSWER 1 OF 1 CAPLUS COPYRIGHT 2001 ACS
ACCESSION NUMBER: 1998:745180 CAPLUS
DOCUMENT NUMBER: 130:21360
TITLE: Nucleic acid molecules specific for bacterial antigens such as O antigens and their sequence and diagnostic and therapeutic uses
INVENTOR(S): Reeves, Peter Richard; Wang, Lei
PATENT ASSIGNEE(S): The University of Sydney, Australia
SOURCE: PCT Int. Appl., 165 pp.
CODEN: PIXXD2
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 1
PATENT INFORMATION

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
WO 9850531	A1	19981112	WO 1998-AU315	19980501
W: AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, CA, CH, CN, CU, CZ, DE, DK, EE, ES, FI, GB, GE, GH, GM, GW, HU, ID, IL, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MD, MG, MK, MN, MW, MX, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, UA, UG, US, UZ, VN, YU, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM				
RW: GH, GM, KE, LS, MW, SD, SZ, UG, ZW, AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, BF, BJ, CF, CG, CI, CM, GA, GN, ML, MR, NE, SN, TD, TG				
AU 9871986	A1	19981127	AU 1998-71986	19980501
EP 1005537	A1	20000607	EP 1998-918970	19980501
R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT, IE, SI, LT, LV, FI, RO				

PRIORITY APPLN. INFO.: AU 1997-6545 19970501
AU 1997-8162 19970722
WO 1998-AU315 19980501

AB The present invention relates to nucleic acid mols. derived from: a gene encoding a transferase; or a gene encoding an enzyme for the transport or processing of a polysaccharide or oligosaccharide unit, including a wzx gene (encoding O antigen flippase) or a wzy gene (encoding O antigen polymerase), or a gene with a similar function; the gene being involved in the synthesis of a particular bacterial polysaccharide antigen, wherein the sequence of the nucleic acid mol. is specific to the particular bacterial polysaccharide antigen. Polysaccharides to which the invention relates include O antigens. The invention also relates to methods of testing samples for the presence of one or more bacterial polysaccharide antigens, using the nucleic acid mols. of the invention, and to kits contg. the nucleic acid mols. of the invention. Thus, gene clusters were sequenced encoding the O antigen synthesis polypeptides from Escherichia coli strains O111 or O157 and from Salmonella enterica (strains C2 or B). The gene sequences and their deduced amino acid sequences are provided, as well as PCR primers designed for the amplification and detection of the genes.

REFERENCE COUNT: 6
REFERENCE(S):
(1) Bastin, D; Gene 1995, V164, P17 CAPLUS
(2) Children'S Hospital And Medical Centre; AU 5391396 A 1996
(3) GOhmann, S; Microbial Pathogenesis 1994, V16, P53 MEDLINE
(4) Luminis Pty Ltd; WO 8912693 1989 CAPLUS
(6) University Of Guelph; WO 9741234 1997 CAPLUS
ALL CITATIONS AVAILABLE IN THE RE FORMAT

L16 ANSWER 1 OF 16 CAPLUS COPYRIGHT 2001 ACS
ACCESSION NUMBER: 1996:649884 CAPLUS
DOCUMENT NUMBER: 125:269274
TITLE: Bacterial GDP **mannose** pyrophosphorylase and GDP-alpha-D-**mannose** manufacture, the preparation of said enzyme and a photometric nucleotidyltransferase assay
INVENTOR(S): Ritter, Joerg Eberhard; Elling, Lothar; Kula, Maria-Regina; Verseck, Stefan
PATENT ASSIGNEE(S) Forschungszentrum Juelich Gmbh, Germany
SOURCE: PCT Int. Appl., 52 pp.
CODEN: PIXXD2
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 1
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
WO 9627670	A2	19960912	WO 1996-DE371	19960301
WO 9627670	A3	19961031		
W: CA, JP, US				
RW: AT, BE, CH, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE				
DE 19606651	A1	19961205	DE 1996-19606651	19960223
CA 2214458	AA	19960912	CA 1996-2214458	19960301
EP 813600	A2	19971229	EP 1996-904730	19960301
R: AT, BE, CH, DE, DK, ES, FR, GB, IT, LI, LU, NL, SE, PT, IE, FI				
JP 11500921	T2	19990126	JP 1996-526527	19960301

PRIORITY APPLN. INFO.: DE 1995-19507449 19950303
DE 1995-19517093 19950515
DE 1996-19606651 19960223
WO 1996-DE371 19960301

AB The invention concerns the gene **rfbM** GDP-**mannose** -pyrophosphorylase from Salmonella enterica. The aim of the invention is to produce a GDP-**mannose**-pyrophosphorylase which can be obtained for an acceptable outlay and does not cause problems, in particular because of its monofunctionality, in continuous multiple stage processes. To that end, a **mannose**- or **mannose**-deriv.-specific GDP-**mannose**-pyrophosphorylase, which can be isolated from microorganisms and has a specific activity of .gtoreq. 2 U/mg, is prepd. The **rfbM** (pyrophosphorylase) and **rfbK** (phosphomannomutase) genes of S. enterica were cloned. The **rfbM** gene was expressed in E. coli and the enzyme purified from the lysate (13.5% yield; 2.34 U/mg). The pyrophosphorylase was partially characterized, e.g. temp. stability, substrate specificity, and km and Vmax. The **rfbK** enzyme was also produced with E. coli and partially purified. Both enzymes were used in prepg. GDP-.alpha.-D-**mannose** from **mannose**. A nucleotidyltransferase assay utilizing pyrophosphate-dependent phosphofructokinase, aldolase, triosephosphate isomerase, and glycerol-3-phosphate dehydrogenase was employed to screen for nucleotidyltransferases in E. coli and rice lysates.

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      (FILE 'HOME' ENTERED AT 16:05:58 ON 02 NOV 2001)

      FILE 'MEDLINE, AGRICOLA, CAPLUS, BIOSIS, EMBASE, WPIDS' ENTERED AT
      16:06:09 ON 02 NOV 2001
L1           714 S PHOSPHOMANNOMUTASE OR (PHOSPHOMANNOSE (W) MUTASE) OR (MANNOSE
L2           841 S L1 OR MANB
L3             2 S L2 AND ((GDP (W) MANNOSE (W) PHOSPHORYLASE) OR (GDP (W) MANN
L4            34 S L2 AND ((GDP (W) MANNOSE (W) PHOSPHORYLASE) OR (GDP (W) MANN
L5            21 DUP REM L4 (13 DUPLICATES REMOVED)
L6             7 S L5 NOT PY>1998

      FILE 'STNGUIDE' ENTERED AT 16:23.36 ON 02 NOV 2001

      FILE 'CAPLUS' ENTERED AT 16:26.52 ON 02 NOV 2001
                E PIEPERSBERG W/AU 25
L7              4 S (E3 OR E4) AND (MANNOSE)
                E DISTLER J/AU 25
L8              3 S (E3 OR E11 OR E12) AND (MANNOSE)
                E ALBERMANN C/AU 25
L9              4 S (E3 OR E4) AND (MANNOSE)
                E TISCHER W/AU 25
L10             0 S (E3 OR E6) AND (MANNOSE)
L11             0 S (E3 OR E6) AND (MANC OR MANB)

      FILE 'STNGUIDE' ENTERED AT 16:37.26 ON 02 NOV 2001

      FILE 'CAPLUS' ENTERED AT 16:48 59 ON 02 NOV 2001
                E JAYARATNE/AU 25
L12             0 S (E9 OR E10) AND (MANC OR MANB)
L13            10 S (E9 OR E10)

      FILE 'STNGUIDE' ENTERED AT 16:57:01 ON 02 NOV 2001

      FILE 'MEDLINE, AGRICOLA, CAPLUS, BIOSIS, EMBASE, WPIDS' ENTERED AT
      17:04:51 ON 02 NOV 2001
L14            60 S RFBK AND RFBM
L15            49 S L14 AND MANNOSE
L16            16 DUP REM L15 (33 DUPLICATES REMOVED)

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L7 ANSWER 1 OF 4 CAPLUS COPYRIGHT 2001 ACS
ACCESSION NUMBER: 2001:592964 CAPLUS
TITLE: Synthesis of the milk oligosaccharide 2'-fucosyllactose using recombinant bacterial enzymes
AUTHOR(S): Albermann, C.; **Piepersberg, W.**; Wehmeier, U. F.
CORPORATE SOURCE: Bergische Universitat GH, Institut fur Chemische Mikrobiologie, Wuppertal, D-42097, Germany
SOURCE: Carbohydr Res. (2001), 334(2), 97-103
CODEN: CRBRAT; ISSN: 0008-6215
PUBLISHER: Elsevier Science Ltd.
DOCUMENT TYPE: Journal
LANGUAGE: English
AB The enzymic synthesis of GDP- beta.-l-fucose and its enzymic transfer reaction using recombinant enzymes from bacterial sources was examd. The GDP-d-**mannose** 4,6-dehydratase and the GDP-4-keto 6-deoxy-d-**mannose** 3,5-epimerase 4-reductase from Escherichia coli K-12, resp., were used to catalyze the conversion of GDP- alpha.-d-**mannose** to GDP-.beta. l fucose with 78% yield. For the transfer of the l-fucose to an acceptor, we cloned and overproduced the .alpha.-(1 2)-fucosyltransferase (FucT2) protein from Helicobacter pylori. We were able to synthesize 2'-fucosyllactose using the overproduced FucT2 enzyme, enzymically synthesized GDP-l-fucose and lactose. The isolation of 2'-fucosyllactose was accomplished by anion-exchange chromatog and gel filtration to give 65% yield.
REFERENCE COUNT: 28
REFERENCE(S): (2) Albermann, C, Glycobiology 2000, V10, P875 CAPLUS
(3) Becker, D; Biochim Biophys Acta 1999, V1455, P193 CAPLUS
(4) Bradford, M; Anal Biochem 1976, V72, P248 CAPLUS
(5) Chan, N; Glycobiology 1995, V5, P683 CAPLUS
(6) Elling, L; Glycobiology 1996, V6, P591 CAPLUS
ALL CITATIONS AVAILABLE IN THE RE FORMAT

L7 ANSWER 2 OF 4 CAPLUS COPYRIGHT 2001 ACS
ACCESSION NUMBER: 2001:591124 CAPLUS
TITLE: Expression and identification of the RfbE protein from Vibrio cholerae O1 and its use for the enzymatic synthesis of GDP-D-perosamine
AUTHOR(S): Albermann, Christoph; **Piepersberg, Wolfgang**
CORPORATE SOURCE: Chemische Mikrobiologie, Bergische Universitat GH Wuppertal, D-42097, Germany
SOURCE: Glycobiology (2001), 11(8), 655-661
CODEN: GLYCE3; ISSN: 0959-6658
PUBLISHER: Oxford University Press
DOCUMENT TYPE: Journal
LANGUAGE: English
AB The 4-amino-6-deoxy-monosaccharide D-perosamine is an important element in the glycosylation of interesting cell products, such as antibiotics and lipopolysaccharides (LPS) of Gram-pos. and Gram-neg. bacteria. The biosynthetic pathway of the precursor mol., GDP-D-perosamine, in Vibrio cholerae O1 starts with an isomerization of fructose-6-phosphate catalyzed by the bifunctional enzyme phosphomannose isomerase-guanosine diphosphomannose pyrophosphorylase (RfbA, E.C. 2.7.7.22) creating the intermediate **mannose**-6-phosphate, which is subsequently converted by the phosphomanno-mutase (RfbB; E.C. 5.4.2.8) and further by RfbA to GDP-D-**mannose**, to GDP-4 keto-6-deoxymannose by a 4,6-dehydratase (RfbD, E.C. 4.2.1.47) and finally to GDP-D-perosamine by an aminotransferase (RfbE; E.C. not yet classified). We cloned the rfbD and the rfbE genes of V. cholerae O1 in Escherichia coli expression vectors. Both biosynthetic enzymes were overproduced in E. coli BL21 (DE3) and their activities were analyzed. The enzymic conversion from GDP-D-**mannose** to GDP-D-perosamine was optimized and the final product, GDP-D-perosamine, was purified and identified by NMR, mass spectrometry, and chromatog. The catalytically active form of the GDP-4-keto-6-deoxy-D-**mannose**-4-aminotransferase seems to be a tetramer of 170 kDa. The His-tag RfbE fusion protein has a Km of 0.06 mM and a Vmax value of 38 nkat/mg protein for the substrate GDP-4-keto-6 deoxy-D-**mannose**. The Km and Vmax values for the cosubstrate L-glutamate were 0.1 mM and 42 nkat/mg protein, resp. The intention of this work is to establish a basis for both the in vitro prodn. of GDP-D-perosamine and for an in vivo perosaminylation system in a suitable bacterial host, preferably E. coli
REFERENCE COUNT: 36
REFERENCE(S): (1) Albermann, C; Glycobiology 2000, V10, P875 CAPLUS
(2) Bilge, S; Infect Immun 1996, V64, P4795 CAPLUS
(3) Bradford, M; Anal Biochem 1976, V72, P248 CAPLUS
(4) Brautaset, T; Chem Biol 2000, V7, P395 CAPLUS
(6) Cloeckaert, A, Res Microbiol 2000, V151, P209 CAPLUS
ALL CITATIONS AVAILABLE IN THE RE FORMAT

L7 ANSWER 3 OF 4 CAPLUS COPYRIGHT 2001 ACS
ACCESSION NUMBER: 2000:656052 CAPLUS
DOCUMENT NUMBER: 134:142489
TITLE: Preparative synthesis of GDP-.beta.-L-fucose by recombinant enzymes from enterobacterial sources
AUTHOR(S): Albermann, Christoph; Distler, Jurgen; **Piepersberg, Wolfgang**
CORPORATE SOURCE: Chemische Mikrobiologie, Bergische Universitat, Wuppertal, D-42097, Germany
SOURCE: Glycobiology (2000), 10(9), 875-881
CODEN: GLYCE3; ISSN: 0959-6658
PUBLISHER: Oxford University Press
DOCUMENT TYPE: Journal
LANGUAGE: English
AB The 6-deoxyhexose L-fucose is an important and characteristic element in glycoconjugates of bacteria (e g., lipopolysaccharides), plants (e.g., xyloglucans) and animals (e.g., glycolipids, glycoproteins, and oligosaccharides). The biosynthetic pathway of GDP-L-fucose starts with a dehydration of GDP-D-**mannose** catalyzed by GDP-D-**mannose** 4,6-dehydratase (Gmd) creating GDP-4-keto-6-deoxymannose which is subsequently converted by the GDP-4-keto-6-deoxy-D-**mannose** 3,5-epimerase-4-reductase (WcaG; GDP-.beta.-L-fucose synthetase) to GDP-.beta.-L-fucose. Both biosynthetic genes gmd and wcaG were cloned from Escherichia coli K12 and the enzymes overexpressed under control of the T7 promoter in the expression vectors pET11a and pET16b, yielding both native and N-terminal His-tag fusion proteins, resp. The activities of the Gmd and WcaG were analyzed. The enzymic conversion from GDP-D-**mannose** to GDP-.beta.-L-fucose was optimized and the final product was purified. The formation of GDP-.beta -L-fucose by the recombinant enzymes was verified by HPLC and NMR analyses. The His-tag fusion variants of the Gmd and WcaG proteins were purified to near homogeneity. The His-tag Gmd recombinant enzyme was inactive, whereas His-tag WcaG showed very similar enzymic properties relative to the native GDP-.beta.-L-fucose synthetase. With the purified His-tag WcaG Km and Vmax values, resp., of 40 .mu.M and 23 nkat/mg protein for the substrate GDP-4-keto-6-deoxy-D-**mannose** and of 21 .mu.M and 10 nkat/mg protein for the cosubstrate NADPH were obtained; a pH optimum of 7.5 was detd. and the enzyme was stimulated to equal extent by the divalent cations Mg2+ and Ca2+. The Gmd enzyme showed a strong feedback inhibition by GDP-.beta.-L-fucose
REFERENCE COUNT: 33
REFERENCE(S): (1) Andrianopoulos, K; J Bacteriol 1998, V180, P998 CAPLUS
(2) Aoyama, K; Mol Biol Evol 1994, V11, P829 CAPLUS
(3) Becker, D; Biochim Biophys Acta 1999, V1455, P193 CAPLUS
(4) Bradford, M; Anal Biochem 1976, V72, P248 CAPLUS
(5) Chan, N; Glycobiol 1995, V5, P683 CAPLUS
ALL CITATIONS AVAILABLE IN THE RE FORMAT

L7 ANSWER 4 OF 4 CAPLUS COPYRIGHT 2001 ACS
ACCESSION NUMBER: 1999:139970 CAPLUS
DOCUMENT NUMBER: 130:195841
TITLE: Method for enzymically producing guanosine diphosphate-6-deoxyhexoses and the use thereof for producing oligosaccharides
INVENTOR(S): **Piepersberg, Wolfgang**; Distler, Jurgen; Albermann, Christoph
PATENT ASSIGNEE(S): Roche Diagnostics G.m.b.H., Germany
SOURCE: PCT Int. Appl., 37 pp.
CODEN: PIXXD2
DOCUMENT TYPE: Patent
LANGUAGE: German
FAMILY ACC. NUM. COUNT: 1
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
WO 9909180	A2	19990225	WO 1998-EP5242	19980818
WO 9909180	A3	19990415		
W: DE, JP, US				
RW: AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE				
DE 19735994	A1	19990225	DE 1997-19735994	19970819
EP 1005554	A2	20000607	EP 1998-943894	19980818
R: AT, BE, CH, DE, DK, ES, FR, GB, IT, LI, NL, SE				
JP 2001514896	T2	20010918	JP 2000-509844	19980818

PRIORITY APPLN. INFO.: DE 1997-19735994 A 19970819
WO 1998-EP5242 W 19980818

AB The invention relates to a method for enzymically prepg. GDP-6-deoxyhexoses from GDP-D-**mannose**, **mannose**

-1-phosphate or **mannose**-6-phosphate in the presence of suitable enzymes, such as a GDP-D-**mannose**-4,6-dehydratase and optionally a GDP-L-fucose synthase or a GDP-4-keto-6-deoxy-D-**mannose** -4-reductase. The invention also relates to a method for coupling the resulting GDP-activated hexoses with oligo- or polysaccharides using glycosyl transferases, e.g., fucosyl transferase.

L6 ANSWER 1 OF 7 MEDLINE
ACCESSION NUMBER: 97294462 MEDLINE
DOCUMENT NUMBER: 97294462 PubMed ID: 9150218
TITLE: Polymorphism, duplication, and IS1-mediated rearrangement in the chromosomal his-rfb-gnd region of Escherichia coli strains with group IA and capsular K antigens.
AUTHOR: Drummelsmith J; Amor P A; Whitfield C
CORPORATE SOURCE: Department of Microbiology, University of Guelph, Ontario, Canada.
SOURCE: JOURNAL OF BACTERIOLOGY, (1997 May) 179 (10) 3232-8.
Journal code: HH3; 2985120R. ISSN 0021-9193.
PUB. COUNTRY: United States
Journal, Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
OTHER SOURCE: GENBANK-U90519
ENTRY MONTH: 199706
ENTRY DATE: Entered STN: 19970620
Last Updated on STN 19990129
Entered Medline: 19970609

AB Individual Escherichia coli strains produce several cell surface polysaccharides. In E. coli E69, the his region of the chromosome contains the rfb (serotype O9 lipopolysaccharide O-antigen biosynthesis) and cps (serotype K30 group IA capsular polysaccharide biosynthesis) loci. Polymorphisms in this region of the Escherichia coli chromosome reflect extensive antigenic diversity in the species. Previously, we reported a duplication of the **manC-manB** genes, encoding enzymes involved in GDP-mannose formation, upstream of rfb in strain E69 (P. Jayaratne et al., J. Bacteriol. 176:3126-3139, 1994). Here we show that one of the **manC-manB** copies is flanked by IS1 elements, providing a potential mechanism for the gene duplication. Adjacent to manB1 on the IS1-flanked segment is a further open reading frame (ugd), encoding uridine-5'-diphosphoglucose dehydrogenase. The Ugd enzyme is responsible for the production of UDP-glucuronic acid, a precursor required for K30 antigen synthesis. Construction of a chromosomal ugd::Gm(r) insertion mutation demonstrated the essential role for Ugd in the biosynthesis of the K30 antigen and confirmed that there is no additional functional ugd copy in strain E69. PCR amplification and Southern hybridization were used to examine the distribution of IS1 elements and ugd genes in the vicinity of rfb in other E. coli strains, producing different group IA K antigens. The relative order of genes and, where present, IS1 elements was established in these strains. The regions adjacent to rfb in these strains are highly variable in both size and gene order, but in all cases where a ugd homolog was present, it was found near rfb. The presence of IS1 elements in the rfb regions of several of these strains provides a potential mechanism for recombination and deletion events which could contribute to the antigenic diversity seen in surface polysaccharides.

L6 ANSWER 2 OF 7 MEDLINE
ACCESSION NUMBER: 96313314 MEDLINE
DOCUMENT NUMBER: 96313314 PubMed ID: 8768520
TITLE: Evidence that the Piromyces gene family encoding endo-1,4-mannanases arose through gene duplication.
AUTHOR: Millward-Sadler S J; Hall J; Black G W; Hazlewood G P; Gilbert H J
CORPORATE SOURCE: Department of Biological and Nutritional Sciences, University of Newcastle upon Tyne, UK.
SOURCE: FEMS MICROBIOLOGY LETTERS, (1996 Aug 1) 141 (2-3) 183-8.
Journal code: FML; 7705721. ISSN: 0378-1097.
PUB. COUNTRY: Netherlands
Journal, Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
OTHER SOURCE: GENBANK-X97408; GENBANK-X97520
ENTRY MONTH: 199612
ENTRY DATE: Entered STN: 19970128
Last Updated on STN 19990129
Entered Medline: 19961205

AB The sequences of two Piromyces cDNAs (**manB** and **manC**) encoding functional mannanases, defined as mannanase B (**MANB**) and mannanase C (**MANC**), revealed that both the cDNAs, and the encoded enzymes, exhibited extensive sequence identity with each other and with a previously described Piromyces mannanase. **MANB** and **MANC**, which belong to glycosyl hydrolase family 26, hydrolyse several forms of mannan but do not attack the other major plant structural polysaccharides. The data presented in this paper indicate that the Piromyces gene family encoding mannanases arose through gene duplication.

L6 ANSWER 3 OF 7 MEDLINE
ACCESSION NUMBER: 91287694 MEDLINE

DOCUMENT NUMBER: 91287694 PubMed ID: 1712067
TITLE: The cps gene cluster of Salmonella strain LT2 includes a second mannose pathway: sequence of two genes and relationship to genes in the rfb gene cluster.
AUTHOR: Stevenson G; Lee S J; Romana L K; Reeves P R
CORPORATE SOURCE: Department of Microbiology, University of Sydney, N.S.W., Australia.
SOURCE: MOLECULAR AND GENERAL GENETICS, (1991 Jun) 227 (2) 173-80. Journal code: NGP; 0125036. ISSN: 0026-8925.
PUB. COUNTRY: GERMANY: Germany, Federal Republic of
Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
OTHER SOURCE: GENBANK-X54103; GENBANK-X59886; GENBANK-X63980, GENBANK-X63981; GENBANK-X63982; GENBANK-X63983, GENBANK-X63984; GENBANK-X63985; GENBANK-X63986, GENBANK-X63987
ENTRY MONTH: 199108
ENTRY DATE: Entered STN 19910825
Last Updated on STN: 19970203
Entered Medline: 19910806

AB We report the presence in Salmonella enterica strain LT2 (serovar thyphimurium) of duplicate genes for two steps in the synthesis of GDP-mannose. The previously known genes, rfbK (**phosphomannomutase**) and rfbM (mannose-1-phosphate guanyltransferase), are part of the gene cluster for the O antigen. The two new genes, cpsB and cpsG, respectively, are thought to be part of the gene cluster for the M antigen capsular polysaccharide, present in many Enterobacteriaceae. The two genes have been sequenced and have a GC content of 0.61, suggesting an origin outside of Salmonella. Comparison of the inferred protein sequences for cpsB and rfbM shows 57% identity of amino acids whereas for cpsG and rfbK there is only 19% identity. It is suggested that the greater divergence between cpsG and rfbK may be due to a period of accelerated evolution, perhaps precipitated by transfer of the genes from another species.

L6 ANSWER 4 OF 7 CAPLUS COPYRIGHT 2001 ACS
ACCESSION NUMBER: 1998.514880 CAPLUS
DOCUMENT NUMBER: 129:226447
TITLE: Organization of Escherichia coli O157 O antigen gene cluster and identification of its specific genes
AUTHOR(S): Wang, Lei; Reeves, Peter R.
CORPORATE SOURCE: Department of Microbiology, The University of Sydney, Sydney, 2006, Australia
SOURCE: Infect. Immun. (1998), 66(8), 3545-3551
CODEN: INFIBR; ISSN: 0019-9567
PUBLISHER: American Society for Microbiology
DOCUMENT TYPE: Journal
LANGUAGE: English

AB The O157:H7 clone of Escherichia coli, which causes major, often prolonged outbreaks of gastroenteritis with hemolytic-uremic syndrome (HUS) such as those in Japan, Scotland, and the United States recently, is thought to be resident normally in cattle or other domestic animals. This clone is of major significance for public health and the food industry. We have developed a fast method for sequencing a given O antigen gene cluster and applied it to O157. The O157 O antigen gene cluster is 14 kb in length, comprising 12 genes and a remnant H-repeat unit. Based on sequence similarity, we have identified all the necessary O antigen genes, including five sugar biosynthetic pathway genes, four transferase genes, the O unit flippase gene, and the O antigen polymerase gene. By PCR testing against all 166 E. coli O serogroups and a range of gram-neg. bacterial strains, including some that cross-react serol. with E. coli O157 antisera, we have found that certain O antigen genes are highly specific to O157 E. coli. This work provides the basis for a sensitive test for rapid detection of O157 E. coli. This is important both for decisions on patient care, since early treatment may reduce the risk of life-threatening complications, and for detection of sources of contamination. The method for fast sequencing of O antigen gene clusters plus an ability to predict which genes will be O antigen specific will enable PCR tests to be developed as needed for other clones of E. coli or, once flanking genes are identified, clones of any gram-neg. bacterium.

L6 ANSWER 5 OF 7 CAPLUS COPYRIGHT 2001 ACS
ACCESSION NUMBER: 1998.301919 CAPLUS
DOCUMENT NUMBER: 129:78994
TITLE: Generation of Escherichia coli O9a serotype, a subtype of E. coli O9, by transfer of the wb* gene cluster of Klebsiella O3 into E. coli via recombination
AUTHOR(S): Sugiyama, Tsuyoshi; Kido, Nobuo; Kato, Yutaka; Koide, Naoki; Yoshida, Tomoaki; Yokochi, Takashi
CORPORATE SOURCE: Department of Microbiology and Immunology and Research Center for Infectious Disease, Aichi Medical University, Nagakute, 480-1195, Japan

SOURCE: J Bacteriol (1998), 180(10), 2775-2778
CODEN: JOBAAY; ISSN: 0021-9193
PUBLISHER: American Society for Microbiology
DOCUMENT TYPE: Journal
LANGUAGE: English
AB Genetic characterization of the wb* gene in a series of Escherichia coli and Klebsiella strains possessing the mannose homopolymer as the O-specific polysaccharide was carried out. The partial nucleotide sequences and PCR-restriction fragment length polymorphism anal. suggested that E. coli serotype O9a, a subtype of E coli O9, might have been generated by the insertion of the Klebsiella O3 wb* gene into a certain E. coli strain.

L6 ANSWER 6 OF 7 CAPLUS COPYRIGHT 2001 ACS
ACCESSION NUMBER: 1997:42445 CAPLUS
DOCUMENT NUMBER: 126:101534
TITLE: Lactobacillus curvatus has a glucose transport system homologous to the mannose family of phosphoenolpyruvate-dependent phosphotransferase systems
AUTHOR(S): Veyrat, Ana; Gosalbes, Maria Jose; Perez-Martinez, Gaspar
CORPORATE SOURCE: Dep. Biotecnologia, Inst. Agroquimica y Tecnologia Alimentos, Valencia, 46100, Spain
SOURCE: Microbiology (Reading, U. K.) (1996), 142(12), 3469-3477
CODEN: MROBEO; ISSN: 1350-0872
PUBLISHER: Society for General Microbiology
DOCUMENT TYPE: Journal
LANGUAGE: English
AB In Lactobacillus curvatus, a phosphoenolpyruvate· mannose phosphotransferase system (mannose-PTS) has been characterized and it was shown to be involved in glucose and mannose transport, but no glucose-specific PTS activity could be detected. A 2 cntdot.1 kb DNA fragment amplified by PCR from the L. curvatus genome was sequenced. Sequence anal. showed four ORFs which could encode proteins similar to PTS transporters EIIA, EIIB, EIIC and EIID of the mannose class. The expression of the **manB** gene (encoding EIIB) from L. curvatus in a mutant of Lactobacillus sake impaired in EIIMan activity restored this activity. Furthermore, this DNA fragment complemented the regulatory function of LevE (EIIB) in a Bacillus subtilis levE-deficient mutant, suggesting that the protein encoded by **manB** could also play a regulatory role in L. curvatus.

L6 ANSWER 7 OF 7 CAPLUS COPYRIGHT 2001 ACS
ACCESSION NUMBER: 1996:494587 CAPLUS
DOCUMENT NUMBER: 125:162882
TITLE: Organization of the Escherichia coli K-12 gene cluster responsible for production of the extracellular polysaccharide colanic acid
AUTHOR(S): Stevenson, Gordon; Andrianopoulos, Kanella; Hobbs, Matthew; Reeves, Peter R.
CORPORATE SOURCE: Dep. Microbiol., Univ Sydney, New South Wales, 2006, Australia
SOURCE: J. Bacteriol. (1996), 178(16), 4885-4893
CODEN: JOBAAY; ISSN: 0021-9193
DOCUMENT TYPE: Journal
LANGUAGE: English
AB Colanic acid (CA) is an extracellular polysaccharide produced by most Escherichia coli strains as well as by other species of the family Enterobacteriaceae. We have detd. the sequence of a 23-kb segment of the E. coli K-12 chromosome which includes the cluster of genes necessary for prodn. of CA. The CA cluster comprises 19 genes. Two other sequenced genes (orf1.3 and galF), which are situated between the CA cluster and the O-antigen cluster, were shown to be unnecessary for CA prodn. The CA cluster includes genes for synthesis of GDP-L-fucose, one of the precursors of CA, and the gene for one of the enzymes in this pathway (GDP-D-mannose 4,6-dehydratase) was identified by biochem. assay. Six of the inferred proteins show sequence similarity to glycosyl transferases, and two others have sequence similarity to acetyl transferases. Another gene (wzx) is predicted to encode a protein with multiple transmembrane segments and may function in export of the CA repeat unit from the cytoplasm into the periplasm in a process analogous to O-unit export. The first three genes of the cluster are predicted to encode an outer membrane lipoprotein, a phosphatase, and an inner membrane protein with an ATP-binding domain. Since homologs of these genes are found in other extracellular polysaccharide gene clusters, they may have a common function, such as export of polysaccharide from the cell.

L8 ANSWER 3 OF 3 CAPLUS COPYRIGHT 2001 ACS
ACCESSION NUMBER 1977:41225 CAPLUS
DOCUMENT NUMBER: 86:41225
TITLE: The role of glycosidically bound **mannose** in the assimilation of .beta.-galactosidase by generalized gangliosidosis fibroblasts
AUTHOR(S): Hieber, Virginia; **Distler, J.**; Myerowitz, Rachel, Schmickel, Roy D.; Jourdian, George W.
CORPORATE SOURCE Dep. Pediatr., Univ. Michigan, Ann Arbor, Mich., USA
SOURCE: Biochem. Biophys. Res. Commun. (1976), 73(3), 710-17
CODEN: BBRCA9
DOCUMENT TYPE: Journal
LANGUAGE: English
AB Bovine testicular .beta.-galactosidase contained equimolar amts. of **mannose** and glucosamine and it strongly bound to concanavalin A-Sepharose Pretreatment of .beta.-galactosidase with a mannosidase prepn. from Aspergillus niger reduced the rate of assimilation of the enzyme 97% in gangliosidosis skin fibroblasts. These data indicated that mannosyl residues play a role in assimilation of the enzyme. This conclusion was supported by obsd. inhibition of .beta.-galactosidase assimilation by **mannose**, methyl .alpha.- and .beta.-mannopyranosides, and **mannose**-contg. testicular glycoproteins.

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L13 ANSWER 7 OF 10 CAPLUS COPYRIGHT 2001 ACS
ACCESSION NUMBER: 1995:899985 CAPLUS
DOCUMENT NUMBER: 124:22965
TITLE: Identification of rcs genes in Escherichia coli O9:K30:H12 and involvement in regulation of expression of group IA K30 capsular polysaccharide
AUTHOR(S): Whitfield, Chris; Keenleyside, Wendy J.; MacLachlan, P. Ronald; **Jayaratne, Padman**; Clarke, Anthony J
CORPORATE SOURCE: Department Microbiology, University Guelph, Guelph, ON, N1G 2W1, Can
SOURCE: Methods Mol. Genet. (1995), 6(Microbial Gene Techniques), 301-22
CODEN: MEMGE6; ISSN: 1067-2389
DOCUMENT TYPE: Journal
LANGUAGE: English
AB Escherichia coli mutants with elevated synthesis of K30 capsular polysaccharide were isolated. The genes involved in regulation of K30 capsular polysaccharide expression were cloned. Cell surface polysaccharides in Escherichia coli K12/K30 hybrids were characterized. Defined mutations in genes rcsA and rcsB of Escherichia coli O9:K30:H12 were constructed. Gene rcs products were required for high level expression of K30 capsular polysaccharide in Escherichia coli. Rcs gene products have a role in regulation of capsular polysaccharide formation in other group K-antigen serotypes.

L13 ANSWER 8 OF 10 CAPLUS COPYRIGHT 2001 ACS
ACCESSION NUMBER: 1994:474960 CAPLUS
DOCUMENT NUMBER: 121:74960
TITLE: Cloning and analysis of duplicated rfbM and rfbK genes involved in the formation of GDP-mannose in Escherichia coli O9:K30 and participation of rfb genes in the synthesis of the group I K30 capsular polysaccharide
AUTHOR(S): **Jayaratne, Padman**; Bronner, Dorothea; MacLachlan, P. Ronald, Dodgson, Christine; Kido, Nobuo; Whitfield, Chris
CORPORATE SOURCE: Dep Microbiol., Univ. Guelph, Ontario, N1G 2W1, Can.
SOURCE: J. Bacteriol (1994), 176(11), 3126-39
CODEN: JOBAAY; ISSN: 0021-9193
DOCUMENT TYPE: Journal
LANGUAGE: English
AB The rfbO9 gene cluster, which is responsible for the synthesis of the lipopolysaccharide O9 antigen, was cloned from Escherichia coli O9:K30. The gnd gene, encoding 6-phosphogluconate dehydrogenase, was identified adjacent to the rfbO9 cluster, and by DNA sequence anal. the gene order gnd-rfbM-rfbK was estd. This order differs from that described for other members of the family Enterobacteriaceae. Nucleotide sequence anal. was used to identify the rfbK and rfbM genes, encoding phosphomannomutase and GDP-mannose pyrophosphorylase, resp. In members of the family Enterobacteriaceae, these enzymes act sequentially to form GDP-mannose, which serves as the activated sugar nucleotide precursor for mannose residues in cell surface polysaccharides. In the E. coli O9:K30 strain, a duplicated rfbM2-rfbK2 region was detected approx. 3 kbp downstream of rfbM1-rfbK1 and adjacent to the remaining genes of the rfbO9 cluster. The rfbM isogenes differed in upstream flanking DNA but were otherwise highly conserved. In contrast, the rfbK isogenes differed in downstream flanking DNA and in 3'-terminal regions, resulting in slight differences in the sizes of the predicted RfbK proteins. RfbMO9 and RfbKO9 are most closely related to CpsB and CpsG, resp. These are isoenzymes of GDP-mannose pyrophosphorylase and phosphomannomutase, resp., which are though to be involved in the biosynthesis of the slime polysaccharide colanic acid in E coli K-12 and Salmonella enterica serovar Typhimurium. An E. coli O-:K30 mutant, strain CWG44, lacks rfbM2-rfbK2 and has adjacent essential rfbO9 sequences deleted The remaining chromosomal genes are therefore sufficient for GDP-mannose formation and K30 capsular polysaccharide synthesis. A mutant of E. coli CWG44, strain CWG152, was found to lack GDP-mannose pyrophosphorylase and lost the ability to synthesize K30 capsular polysaccharide. Wild-type capsular polysaccharide could be restored in CWG152, by transformation with plasmids contg. either rfbM1 or rfbM2. Introduction of a complete rfbO9 gene cluster into CWG152 restored synthesis of both O9 and K30 polysaccharides. Consequently, rfbM is sufficient for the biosynthesis of GDP-mannose for both O antigen and capsular polysaccharide in E coli O9:K30 Anal. of a collection of serotype O8 and O9 isolates by Southern hybridization and PCR amplification expts. demonstrated extensive polymorphism in the rfbM-rfbK region.

L13 ANSWER 9 OF 10 CAPLUS COPYRIGHT 2001 ACS
ACCESSION NUMBER: 1994:1842 CAPLUS

DOCUMENT NUMBER: 120:1842
TITLE: Characterization of rcsB and rcsC from Escherichia coli O9:K30:H12 and examination of the role of the rcs regulatory system in expression of group I capsular polysaccharides
AUTHOR(S): **Jayaratne, Padman**; Keenleyside, Wendy J.; MacLachlan, P. Ronald; Dodgson, Christine; Whitfield, Chris
CORPORATE SOURCE: Dep. Microbiol., Univ. Guelph, Guelph, ON, N1G 2W1, Can.
SOURCE: J. Bacteriol. (1993), 175(17), 5384-94
CODEN: JOBAAY; ISSN: 0021-9193
DOCUMENT TYPE: Journal
LANGUAGE: English

AB In Escherichia coli K-12, RcsD and RcsB are thought to act as the sensor and effector components, resp., of a two-component regulatory system which regulates expression of the slime polysaccharide colanic acid (V. Stout and S. Gottesman, 1990). Here, the authors report the cloning and DNA sequence of a 4.3-kb region contg. rcsC and rcsB from E. coli O9:K30:H12. This strain does not produce colanic acid but does synthesize a K30 (group I) capsular polysaccharide. The rcsB gene from E. coli K30 (rcsBK30) is identical to the rcsB gene from E. coli K-12 (rcsBK-12). RcsCK30 has 16 nucleotide changes, resulting in six amino acid changes in the predicted protein. To examine the function of the rcs regulatory system in expression of the K30 capsular polysaccharide, chromosomal insertion mutations were constructed in E. coli O9:K30:H12 to independently inactivate rcsBK30 and the auxiliary pos. regulator rcsAK30. Strains with these mutations maintained wild-type levels of K30 capsular polysaccharide expression and still produced a K30 capsule, indicating that the rcs system is not essential for expression of low levels of the group I capsular polysaccharide in lon+ E. coli K30. However, K30 synthesis is increased by introduction of a multicopy plasmid carrying rcsBK30. K30 polysaccharide expression is also markedly elevated in an rcsBK30-dependent fashion by a mutation in rcsCK30, suggesting that the rcs system is involved in high levels of synthesis. To det. whether the involvement of the rcs system in E. coli K30 expression is typical of group I (K antigen) capsules, multicopy rcsBK30 was introduced into 22 addnl. strains with structurally different group I capsules. All showed an increase in mucoid phenotype, and the polysaccharides produced in the presence and absence of multicopy rcsBK30 were examd. It is has been suggested that E. coli strains with group I capsules can be subdivided based on K antigen structure. For the first time, the authors show that strains with group I capsules can also be subdivided by the ability to produce colanic acid. Group IA contains capsular polysaccharides (including K30) with repeating-unit structures lacking amino sugars, and expression of group IA capsular polysaccharides is increased by multicopy rcsBK30. Group IB capsular polysaccharides all contain amino sugars. In group IB strains, multicopy rcsBK30 activates synthesis of colanic acid.

L13 ANSWER 10 OF 10 CAPLUS COPYRIGHT 2001 ACS
ACCESSION NUMBER: 1993:17265 CAPLUS
DOCUMENT NUMBER: 118:17265
TITLE: The rcsA gene of Escherichia coli O9:K30H12 is involved in the expression of the serotype-specific group I K (capsular) antigen
AUTHOR(S): Keenleyside, Wendy J.; **Jayaratne, Padman**; MacLachlan, P. Ronald; Whitfield, Chris
CORPORATE SOURCE: Dep. Microbiol., Univ. Guelph, Guelph, ON, N1G 2W1, Can.
SOURCE: J. Bacteriol. (1992), 174(1), 8-16
CODEN: JOBAAY; ISSN: 0021-9193
DOCUMENT TYPE: Journal
LANGUAGE: English

AB E. coli produces 2 distinct types of capsular polysaccharide (designated groups I and II), which are distinguished by chem., phys., and genetic characteristics. The K30 capsular antigen is a member of the group I, or heat-stable, capsules. Gene rcsA was cloned from E. coli O9:K30 and its nucleotide sequence was detd. The rcsAK30 sequence is virtually identical to the rcsAK-12 sequence (Stout, V., et al., 1991). RcsAK-12 is a transcriptional activator involved in expression of the extracellular polysaccharide colanic acid in E. coli K-12. rcsAK30 Complemented an rcsAK-12 mutation and activated colanic acid synthesis in E. coli K-12 strains. However, in E. coli K30, increasing the levels of RcsA by introducing multicopy rcsAK30 or a Lon mutation resulted in elevated synthesis of the K30 capsular polysaccharide; no colanic acid was detected. E. coli K-12 strains in which the chromosomal his region was replaced by that from E. coli K30 were able to synthesize K30 capsular polysaccharide. These K-12/K30 hybrid strains did not produce colanic acid, suggesting that the genes for synthesis of colanic acid and the K30 capsular polysaccharide may be allelic. rcsA Sequences were also detected in the group II strains E. coli K1 and K5. Introduction of rcsAK30 into group II strains resulted in activation of colanic acid biosynthesis

rather than the group II capsule. Given the role of RcsA in other members of the family Enterobacteriaceae, these results provide further evidence that this protein may be a relatively widespread regulatory component for the synthesis of enterobacterial extracellular polysaccharides.

UE? Y/(N) :y

L16 ANSWER 1 OF 16 CAPLUS COPYRIGHT 2001 ACS
ACCESSION NUMBER: 1996 649884 CAPLUS
DOCUMENT NUMBER: 125:269274
TITLE: Bacterial GDP **mannose** pyrophosphorylase and GDP-alpha-D-**mannose** manufacture, the preparation of said enzyme and a photometric nucleotidyltransferase assay
INVENTOR(S): Ritter, Joerg Eberhard; Elling, Lothar; Kula, Maria-Regina; Verseck, Stefan
PATENT ASSIGNEE(S): Forschungszentrum Juelich Gmbh, Germany
SOURCE: PCT Int. Appl., 52 pp.
CODEN: PIXXD2
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 1
PATENT INFORMATION

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
WO 9627670	A2	19960912	WO 1996-DE371	19960301
WO 9627670	A3	19961031		
W: CA, JP, US				
RW: AT, BE, CH, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE				
DE 19606651	A1	19961205	DE 1996-19606651	19960223
CA 2214458	AA	19960912	CA 1996-2214458	19960301
EP 813600	A2	19971229	EP 1996-904730	19960301
R: AT, BE, CH, DE, DK, ES, FR, GB, IT, LI, LU, NL, SE, PT, IE, FI				
JP 11500921	T2	19990126	JP 1996-526527	19960301

PRIORITY APPLN. INFO.:
DE 1995-19507449 19950303
DE 1995-19517093 19950515
DE 1996-19606651 19960223
WO 1996-DE371 19960301

AB The invention concerns the gene **rfbM** GDP-**mannose** -pyrophosphorylase from Salmonella enterica. The aim of the invention is to produce a GDP-**mannose**-pyrophosphorylase which can be obtained for an acceptable outlay and does not cause problems, in particular because of its monofunctionality, in continuous multiple stage processes. To that end, a **mannose**- or **mannose**-deriv.-specific GDP-**mannose**-pyrophosphorylase, which can be isolated from microorganisms and has a specific activity of .gtoreq. 2 U/mg, is prepd. The **rfbM** (pyrophosphorylase) and **rfbK** (phosphomannomutase) genes of S. enterica were cloned. The **rfbM** gene was expressed in E. coli and the enzyme purified from the lysate (13.5% yield; 2.34 U/mg). The pyrophosphorylase was partially characterized, e.g. temp stability, substrate specificity, and km and Vmax. The **rfbK** enzyme was also produced with E. coli and partially purified. Both enzymes were used in prepg. GDP-.alpha.-D-**mannose** from **mannose**. A nucleotidyltransferase assay utilizing pyrophosphate-dependent phosphofructokinase, aldolase, triosephosphate isomerase, and glycerol-3-phosphate dehydrogenase was employed to screen for nucleotidyltransferases in E. coli and rice lysates

L16 ANSWER 2 OF 16 MEDLINE DUPLICATE 1
ACCESSION NUMBER: 97081711 MEDLINE
DOCUMENT NUMBER: 97081711 PubMed ID: 8922954
TITLE: Expression, purification and characterization of recombinant phosphomannomutase and GDP-alpha-D-**mannose** pyrophosphorylase from Salmonella enterica, group B, for the synthesis of GDP-alpha-D-**mannose** from D-**mannose**.
AUTHOR: Elling L; Ritter J E; Verseck S
CORPORATE SOURCE: Institut fur Enzymtechnologie, Heinrich-Heine-Universitat DAsseldorf im Forschungszentrum Julich, Germany.
SOURCE: GLYCOBIOLOGY, (1996 Sep) 6 (6) 591-7.
Journal code: BEL, 9104124. ISSN: 0959-6658.
PUB. COUNTRY: ENGLAND: United Kingdom
Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 199702
ENTRY DATE: Entered STN: 19970305
Last Updated on STN 19970305
Entered Medline 19970218

AB The genes **rfbK** and **rfbM** from the rfb cluster (O antigen biosynthesis) of Salmonella enterica, group B, encoding for the enzymes phosphomannomutase (EC 5 4.2.8) and GDP-alpha-D-**mannose** pyrophosphorylase (EC 2.7.7.13) were overexpressed in E.coli BL21 (DE3) with specific activities of 0.1 U/mg and 0.3-0.6 U/mg, respectively. Both enzymes were partially purified to give specific activities of 0.26 U/mg

and 2.75 U/mg, respectively. Kinetic characterization of the homodimeric (108 kDa) GDP-alpha-D-**mannose** pyrophosphorylase revealed a K(m) for GTP and **mannose**-1-P of 0.2 mM and 0.01 mM with substrate surplus inhibition constants (Kis) of 10.9 mM and 0.7 mM, respectively. The product GDP-alpha D **mannose** gave a competitive inhibition with respect to GTP (Ki 14 7 microM) and an uncompetitive inhibition with respect to **mannose**-1-P (Ki 115 microM). Both recombinant enzymes were used for repetitive batch synthesis of GDP-alpha-D-**mannose** staring from D-**mannose** and GTP. In three subsequent batches 581 mg (960 mumol) GDP-alpha-D-**mannose** was synthesized with 80% average yield. The overall yield after product isolation was 22.9% (329 mumol, 199 mg).

L16 ANSWER 3 OF 16 MEDLINE DUPLICATE 2
ACCESSION NUMBER: 97086507 MEDLINE
DOCUMENT NUMBER: 97086507 PubMed ID: 8932701
TITLE: The gene cluster directing O-antigen biosynthesis in Yersinia enterocolitica serotype O:8: identification of the genes for **mannose** and galactose biosynthesis and the gene for the O-antigen polymerase.
AUTHOR: Zhang L; Toivanen P; Skurnik M
CORPORATE SOURCE: Department of Medical Microbiology, University of Turku, Finland.. Lzhang@finabo.abo.fi
SOURCE: MICROBIOLOGY, (1996 Feb) 142 (Pt 2) 277-88.
Journal code: BXW; 9430468. ISSN: 1350-0872.
PUB. COUNTRY: ENGLAND. United Kingdom
Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
OTHER SOURCE: GENBANK-U46859
ENTRY MONTH: 199612
ENTRY DATE: Entered STN: 19970128
Last Updated on STN: 19980206
Entered Medline: 19961231

AB The rfb gene cluster of Yersinia enterocolitica serotype O:8 (YeO8) strain 8081-c was cloned by cosmid cloning. Restriction mapping, deletion analysis and transposon mutagenesis showed that about 19 kb of the cloned DNA is essential for the synthesis and expression of the YeO8 O-side-chain in Escherichia coli Deletion analysis generated a derivative that expressed semi-rough LPS, a phenotype typical of an rfc mutant lacking the O-antigen polymerase. The deletions and transcomplementation experiments allowed localization of the rfc gene to the 3'-end of the rfb gene cluster The deduced YeO8 Rfc did not share significant amino acid sequence similarity with any other protein, but its amino acid composition and hydrophobicity profile are similar to those of identified Rfc proteins. In addition, the codon usage of the rfc gene is similar to other rfc genes. Nucleotide sequence analysis identified three other genes upstream of rfc. Two of the gene products showed 60-70% identity to the **RfbM** and **RfbK** proteins that are biosynthetic enzymes for the GDPmannose pathway of enterobacteria. The third gene product was about 50-80% identical to the bacterial GalE protein, UDPglucose 4-epimerase, which catalyses the epimerization of UDPglucose to UDPgalactose. Since **mannose** and galactose are both present in the YeO8 O-antigen repeat unit, the above three genes are likely to belong to the rfb gene cluster. A gene similar to the gsk gene downstream of rfc, and genes similar to adk and hemH upstream of the rfb gene cluster, were recognized. Thus the rfb gene cluster of YeO8 is located between the adk-hemH and gsk loci, and the order is adk-hemH-rfb-rfc-gsk in the chromosome. Also in other Yersinia spp., the locus downstream of the hemH gene is occupied by gene clusters associated with LPS biosynthesis.

L16 ANSWER 4 OF 16 MEDLINE DUPLICATE 3
ACCESSION NUMBER: 95238291 MEDLINE
DOCUMENT NUMBER: 95238291 PubMed ID: 7536735
TITLE: Expression of the O9 polysaccharide of Escherichia coli: sequencing of the E. coli O9 rfb gene cluster, characterization of mannosyl transferases, and evidence for an ATP-binding cassette transport system.
AUTHOR: Kido N; Torgov V I; Sugiyama T; Uchiya K; Sugihara H; Komatsu T; Kato N; Jann K
CORPORATE SOURCE: Max-Planck-Institute fur Immunobiologie, Freiburg, Germany.
SOURCE: JOURNAL OF BACTERIOLOGY, (1995 Apr) 177 (8) 2178-87.
Journal code HH3; 2985120R. ISSN 0021-9193.
PUB. COUNTRY: United States
Journal, Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
OTHER SOURCE: GENBANK-D43637
ENTRY MONTH: 199505
ENTRY DATE: Entered STN: 19950605
Last Updated on STN: 19960129
Entered Medline: 19950519

AB The rfb gene cluster of Escherichia coli O9 directs the synthesis of the O9-specific polysaccharide which has the structure -->2-alpha-Man-(1-->2)-alpha-Man-(1-->2)-alpha-Man-(1-->3)-alpha- Man-(1--> The E. coli O9 rfb cluster has been sequenced, and six genes, in addition to the previously described **rfbK** and **rfbM**, were identified. They correspond to six open reading frames (ORFs) encoding polypeptides of 261, 431, 708, 815, 381, and 274 amino acids. They are all transcribed in the counter direction to those of the his operon No gene was found between rfb and his. A higher G+C content indicated that E. coli O9 rfb evolved independently of the rfb clusters from other E. coli strains and from Shigella and Salmonella spp. Deletion mutagenesis, in combination with analysis of the in vitro synthesis of the O9 mannan in membranes isolated from the mutants, showed that three genes (termed mtfA, -B, and -C, encoding polypeptides of 815, 381, and 274 amino acids, respectively) directed alpha-mannosyl transferases. MtfC (from ORF274), the first mannosyl transferase, transfers a **mannose** to the endogenous acceptor. It critically depended on a functional rfe gene (which directs the synthesis of the endogenous acceptor) and initiates the growth of the polysaccharide chain. MtfB (from ORF381) then transfers two mannoses into the 3 position of the previous **mannose**, and MtfA (from ORF815) transfers three mannoses into the 2 position Further chain growth needs only the two transferases MtfA and MtfB Thus, there are fewer transferases needed than the number of sugars in the repeating unit.(ABSTRACT TRUNCATED AT 250 WORDS)

L16 ANSWER 5 OF 16 MEDLINE
ACCESSION NUMBER: 96105197 MEDLINE
DOCUMENT NUMBER: 96105197 PubMed ID: 8529890
TITLE: A putative pathway for biosynthesis of the O-antigen component, 3-deoxy-L-glycero-tetronic acid, based on the sequence of the Vibrio cholerae O1 rfb region.
AUTHOR: Morona R; Stroeher U H; Karageorgos L E; Brown M H; Manning P A
CORPORATE SOURCE: Department of Microbiology and Immunology, University of Adelaide, Australia.
SOURCE: GENE, (1995 Dec 1) 166 (1) 19-31.
Journal code FOP; 7706761 ISSN: 0378-1119.
PUB. COUNTRY: Netherlands
Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
OTHER SOURCE: GENBANK-A32047, GENBANK-D00938; GENBANK-D13262; GENBANK-M93187, GENBANK-P02901; GENBANK-P06758; GENBANK-P08659, GENBANK-P09095; GENBANK-P11549; GENBANK-P12784, GENBANK-P12884; GENBANK-P13129; GENBANK-P13604, GENBANK-P14688; GENBANK-P14912; GENBANK-P15727, GENBANK-P16928; GENBANK-P17814; GENBANK-P19372, GENBANK-P25464; GENBANK-P27742; GENBANK-P29212, GENBANK-P31552; GENBANK-S26421; GENBANK-X03721, GENBANK-X04379; GENBANK-X15577; GENBANK-X16144; GENBANK-X59553; GENBANK-X59554; +
ENTRY MONTH: 199601
ENTRY DATE: Entered STN: 19960220
Last Updated on STN: 19960220
Entered Medline: 19960126

AB The nucleotide sequence of a region of the rfb genes, encoding biosynthesis of the Vibrio cholerae (Vc) O1 O-antigen, was determined. Analysis of the open reading frames (ORFs) within this region has revealed similarities with a number of different classes of biosynthetic proteins and enzymes. The ORFs have been designated **RfbK**, RfbL, **RfbM**, RfbN and RfbO. **RfbK** is a small, acidic protein which has similarity to the family of proteins known as acyl-carrier proteins (ACP). The RfbL protein has similarity to a super-family of enzymes which adenylate their substrates as a part of their reaction mechanism. Included in these are several acetyl-CoA ligases. Alignment of RfbL with these proteins reveals a highly conserved domain containing the motif GlyXaaXaaGlyXaaPro. This resembles the ATP-binding site motif and may represent a variant of the usual motif, except that Pro replaces Gly. The VcRfbM protein has similarity with a family of long-chain, iron-containing alcohol dehydrogenases, of which the Escherichia coli K-12 fucO and adhE gene products are also members The RfbN protein has sequence homology with LuxE and LuxC of Vibrio harveyi (Vh) and other bioluminescent bacterial species. The latter are two components of the enzyme complex which synthesizes the long-chain aldehyde used in the V. harveyi bioluminescence system. Finally, the VcRfbO protein has sequence similarity with acetyl-CoA transferases. We were able to identify a number of the gene products using a T7 expression system, confirming several of the allocated ORFs. A biosynthetic pathway for the Vc O-antigen component 3-deoxy-L-glycero-tetronic acid, based on the enzymatic functions predicted for the **RfbK**, RfbL, **RfbM**, RfbN and RfbO proteins, is presented

L16 ANSWER 6 OF 16 MEDLINE DUPLICATE 4
ACCESSION NUMBER: 96060831 MEDLINE
DOCUMENT NUMBER: 96060831 PubMed ID: 7590310
TITLE: Sequence and analysis of the O antigen gene (rfb) cluster of Escherichia coli O111.
AUTHOR: Bastin D A; Reeves P R
CORPORATE SOURCE: Department of Microbiology, University of Sydney, New South Wales, Australia.
SOURCE: GENE, (1995 Oct 16) 164 (1) 17-23.
Journal code: FOP; 7706761. ISSN: 0378-1119.
PUB. COUNTRY: Netherlands
Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
OTHER SOURCE: GENBANK-U13629
ENTRY MONTH: 199512
ENTRY DATE: Entered STN: 19960124
Last Updated on STN: 19960124
Entered Medline: 19951212

AB The O antigens found in Salmonella enterica (Se) and Escherichia coli (Ec) show a great deal of diversity, and only three structures are known to be common to both genera. Two of them contain the 3,6-dideoxyheoxse colitose, not found in other serogroups of the two species. The first of these is common to Ec O111 and Se O:35 (sv Adelaide), the other is found in both Ec O55 and Se O:50 (sv Greenside). The genes specific for the synthesis of O antigen are generally located in the rfb gene cluster at map position 45 min in Ec and 42 min in Se. The rfb (O antigen) gene cluster of an Ec O111 strain M92 had been cloned earlier and hybridisation analysis suggested that the rfb clusters of Ec M92 and a Se sv Adelaide strain had been acquired separately by the two species since their divergence. We have now sequenced part of the rfb cluster from Ec M92. We identify two genes of the GDP-colitose pathway, **rfbM** and **rfbK**, and show that several other ORFs have similarity to the rfb and cps (capsular polysaccharide) genes. Downstream of this block of genes is an ORF which encodes a protein with predicted transmembrane segments which is presumed to correspond to the rfbX gene. The % G+C values of the Ec M92 rfb sequence are extremely low, indicating that the rfb evolved in a low % G+C species of bacteria before transfer into Ec.

L16 ANSWER 7 OF 16 MEDLINE DUPLICATE 5
ACCESSION NUMBER: 94252978 MEDLINE
DOCUMENT NUMBER: 94252978 PubMed ID: 7515042
TITLE: Cloning and analysis of duplicated **rfbM** and **rfbK** genes involved in the formation of GDP-**mannose** in Escherichia coli O9:K30 and participation of rfb genes in the synthesis of the group I K30 capsular polysaccharide.
AUTHOR: Jayaratne P; Bronner D; MacLachlan P R; Dodgson C; Kido N; Whitfield C
CORPORATE SOURCE: Department of Microbiology, University of Guelph, Ontario, Canada.
SOURCE: JOURNAL OF BACTERIOLOGY, (1994 Jun) 176 (11) 3126-39.
Journal code: HH3; 2985120R. ISSN: 0021-9193.
PUB. COUNTRY: United States
Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
OTHER SOURCE: GENBANK-L27632; GENBANK-L27646
ENTRY MONTH: 199406
ENTRY DATE: Entered STN: 19940707
Last Updated on STN: 19960129
Entered Medline: 19940630

AB The rfbO9 gene cluster, which is responsible for the synthesis of the lipopolysaccharide O9 antigen, was cloned from Escherichia coli O9:K30. The gnd gene, encoding 6-phosphogluconate dehydrogenase, was identified adjacent to the rfbO9 cluster, and by DNA sequence analysis the gene order gnd-**rfbM**-**rfbK** was established. This order differs from that described for other members of the family Enterobacteriaceae. Nucleotide sequence analysis was used to identify the **rfbK** and **rfbM** genes, encoding phosphomannomutase and GDP-**mannose** pyrophosphorylase, respectively. In members of the family Enterobacteriaceae, these enzymes act sequentially to form GDP-**mannose**, which serves as the activated sugar nucleotide precursor for **mannose** residues in cell surface polysaccharides. In the E. coli O9:K30 strain, a duplicated rfbM2-rfbK2 region was detected approximately 3 kbp downstream of rfbM1-rfbK1 and adjacent to the remaining genes of the rfbO9 cluster. The **rfbM** isogenes differed in upstream flanking DNA but were otherwise highly conserved. In contrast, the **rfbK** isogenes differed in downstream flanking DNA and in 3'-terminal regions, resulting in slight differences in the sizes of the predicted **RfbK** proteins. RfbMO9 and RfbKO9 are most closely related to CpsB and CpsG, respectively. These are isozymes of GDP-

mannose pyrophosphorylase and phosphomannomutase, respectively, which are thought to be involved in the biosynthesis of the slime polysaccharide colanic acid in E. coli K-12 and Salmonella enterica serovar Typhimurium. An E. coli O-:K30 mutant, strain CWG44, lacks rfbM2-rfbK2 and has adjacent essential rfbO9 sequences deleted. The remaining chromosomal genes are therefore sufficient for GDP-**mannose** formation and K30 capsular polysaccharide synthesis. A mutant of E. coli CWG44, strain CWG152, was found to lack GDP-**mannose** pyrophosphorylase and lost the ability to synthesize K30 capsular polysaccharide. Wild-type capsular polysaccharide could be restored in CWG152, by transformation with plasmids containing either rfbM1 or rfbM2. Introduction of a complete rfbO9 gene cluster into CWG152 restored synthesis of both O9 and K30 polysaccharides. Consequently, **rfbM** is sufficient for the biosynthesis of GDP-**mannose** for both O antigen and capsular polysaccharide E coli O9:K30. Analysis of a collection of serotype O8 and O9 isolates by Southern hybridization and PCR amplification experiments demonstrated extensive polymorphism in the **rfbM-rfbK** region.

L16 ANSWER 8 OF 16 MEDLINE DUPLICATE 6
ACCESSION NUMBER: 94214678 MEDLINE
DOCUMENT NUMBER: 94214678 PubMed ID: 8162191
TITLE: Genetic analysis of Escherichia coli O9 rfb: identification and DNA sequence of phosphomannomutase and GDP-**mannose** pyrophosphorylase genes.
AUTHOR: Sugiyama T; Kido N; Komatsu T; Ohta M; Jann K; Jann B; Saeki A; Kato N
CORPORATE SOURCE: Department of Bacteriology, Nagoya University School of Medicine, Aichi, Japan.
SOURCE: MICROBIOLOGY, (1994 Jan) 140 (Pt 1) 59-71.
Journal code: BXW; 9430468. ISSN: 1350-0872
PUB. COUNTRY: ENGLAND: United Kingdom
Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
OTHER SOURCE: GENBANK-D43637
ENTRY MONTH: 199405
ENTRY DATE: Entered STN: 19940606
Last Updated on STN: 19990129
Entered Medline: 19940520

AB Subcloning, transposon insertion, and deletion analysis revealed that the Escherichia coli O9 rfb region is about 12 kb in size. The region encodes at least seven polypeptides of 89, 74, 55, 50, 44, 41 and 39.5 kDa. Southern hybridization analysis of rfb regions of E. coli O8 and O9, and Klebsiella O3 and O5 serotypes (all of these O polysaccharides are **mannose** homopolymers and the structures of the repeating unit of E. coli O9 and Klebsiella O3 are identical) showed that a central region specific for E. coli O9 and Klebsiella O3 is flanked by two regions common to all four. Complementation experiments using strains with known defects and specific tests for the enzymic activity showed that the 50 and 55 kDa polypeptides, encoded by the common region, are phosphomannomutase (PMM) and GDP-**mannose** pyrophosphorylase (GMP), respectively. Nucleotide sequencing of the region revealed the presence of two genes, **rfbK** and **rfbM**, analogous to the corresponding genes of Salmonella typhimurium. In E. coli O9, **rfbK** and **rfbM** encode proteins of 460 amino acids (50,809 Da) and 471 amino acids (52,789 Da). The amino acid sequence of GMP was conserved in RfbMs of E. coli O7 and Salmonella groups B, C1 and C2, CpsB of S. typhimurium, AlgA of Pseudomonas aeruginosa, and XanB of Xanthomonas campestris. The phylogenetic trees of PMM and GMP were different in topology and in the evolutionary distances from ancestors.

L16 ANSWER 9 OF 16 MEDLINE DUPLICATE 7
ACCESSION NUMBER: 93328694 MEDLINE
DOCUMENT NUMBER: 93328694 PubMed ID: 7687601
TITLE: Variation of the rfb gene clusters in Salmonella enterica.
AUTHOR: Xiang S H; Haase A M; Reeves P R
CORPORATE SOURCE: Department of Microbiology, University of Sydney, New South Wales, Australia.
SOURCE: JOURNAL OF BACTERIOLOGY, (1993 Aug) 175 (15) 4877-84.
Journal code: HH3; 2985120R ISSN: 0021-9193
PUB. COUNTRY: United States
Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 199308
ENTRY DATE: Entered STN: 19930903
Last Updated on STN: 19980206
Entered Medline: 19930825

AB In order to explore the genetic variation of O antigens of Salmonella enterica, we surveyed 164 strains (132 serovars) belonging to 45 serogroups, using 25 mostly single-gene rfb DNA probes for colony

hybridization. The results revealed that strains within a serogroup have very similar or identical rfb genes. At least three of the four rhamnose genes were detected in all 17 serogroups reported to contain rhamnose, and one or more were detected in three others. The likelihood of being detected decreased in the order rfbB, rfbC, rfbA, and rfbD, which is the map order, suggesting a gradient of divergence. **Mannose** pathway genes were much less conserved, and of 27 groups reported to contain **mannose** or **mannose** derivatives colitose or fucose, only 9 hybridized to the **rfbM** and **rfbK** probes. Dideoxyhexose genes were found only in groups reported to contain dideoxyhexoses. Group D2, which had not been studied previously, appears to resemble group D1, with the substitution of one gene from group E1 to give a change in one linkage. In contrast to sugar pathway genes, sugar transferase genes did not in general hybridize to strains of other groups outside the closely related groups A, B, and D, with the exception of the galactose transferase gene also shared by groups C2, C3, and all E groups.

L16 ANSWER 10 OF 16 CAPLUS COPYRIGHT 2001 ACS DUPLICATE 8
ACCESSION NUMBER: 1993:487831 CAPLUS
DOCUMENT NUMBER: 119:87831
TITLE: Identification, expression, and DNA sequence of the GDP-**mannose** biosynthesis genes encoded by the O7 rfb gene cluster of strain VW187 (Escherichia coli O7:K1)
AUTHOR(S): Marolda, Cristina L.; Valvano, Miguel A.
CORPORATE SOURCE: Dep. Microbiol. Immunol., Univ. West. Ontario, London, ON, N6A 5C1, Can.
SOURCE: J. Bacteriol. (1993), 175(1), 148-58
CODEN: JOBAAY; ISSN: 0021-9193
DOCUMENT TYPE: Journal
LANGUAGE: English
AB The O7-specific lipopolysaccharide (LPS) in strains of E. coli consists of a repeating unit made of galactose, **mannose**, rhamnose, 4-acetamido-2,6-dideoxyglucose, and N-acetylglucosamine. The O7-specific LPS biosynthesis region (rfbEcO7) of the E. coli O7:K1 strain VW187 was recently cloned and characterized genetically . In this study, the gnd gene encoding gluconate-6-phosphate dehydrogenase was localized at one end of the rfbEcO7 gene cluster and that end of the cluster was sequenced. Three open reading frames (ORF) encoding polypeptides of 275, 464, and 453 amino acids were identified upstream of gndEcO7, all transcribed toward the gnd gene. ORF275 had 45% similarity at the protein level with ORF16.5, which occupies a similar position in the Salmonella enterica LT2 rfb region, and presumably encodes a nucleotide sugar transferase. The polypeptides encoded by ORFs 464 and 453 were expressed under the control of the ptac promoter and visualized in Coomassie blue-stained SDS gels and by maxicell anal. ORF464 expressed GDP-**mannose** pyrophosphorylase and ORF453 encoded a phosphomannomutase, the enzymes for the biosynthesis pathway of GDP-**mannose**, one of the nucleotide sugar precursors for the formation of the O7 repeating unit. They were designated rfbMEcO7 and rfbKEcO7, resp. The RfbMEcO7 polypeptide was homologous to the corresponding protein in S. enterica LT2, XanB of Xanthomonas campestris, and AlgA of Pseudomonas aeruginosa, all GDP-**mannose** pyrophosphorylases. RfbKEcO7 was very similar to CpsG of S. enterica LT2, an enzyme presumably involved in the biosynthesis of the capsular polysaccharide colanic acid, but quite different from the corresponding **RfbK** protein of S. enterica LT2.

L16 ANSWER 11 OF 16 BIOSIS COPYRIGHT 2001 BIOSIS
ACCESSION NUMBER: 1993:357580 BIOSIS
DOCUMENT NUMBER: PREV199345041005
TITLE: The **rfbM** and **rfbK** gene products are shared in biosynthesis of the 09 and K30 polysaccharides in Escherichia coli 09:K30.
AUTHOR(S): Jayaratne, P.; Maclachlan, P. R.; Whitfield, C.
CORPORATE SOURCE: Univ. Guelph, Guelph, ON N1G 2W1 Canada
SOURCE: Abstracts of the General Meeting of the American Society for Microbiology, (1993) Vol. 93, No. 0, pp. 138.
Meeting Info.: 93rd General Meeting of the American Society for Microbiology Atlanta, Georgia, USA May 16-20, 1993
ISSN: 1060-2011
DOCUMENT TYPE: Conference
LANGUAGE: English

L16 ANSWER 12 OF 16 MEDLINE DUPLICATE 9
ACCESSION NUMBER: 93018988 MEDLINE
DOCUMENT NUMBER: 93018988 PubMed ID: 1383393
TITLE: Sequence and structural analysis of the rfb (O antigen) gene cluster from a group C1 Salmonella enterica strain.
AUTHOR: Lee S J; Romana L K; Reeves P R
CORPORATE SOURCE: Department of Microbiology, University of Sydney, Australia.
SOURCE: JOURNAL OF GENERAL MICROBIOLOGY, (1992 Sep) 138 (Pt 9)

1843-55.
Journal code: I87; 0375371 ISSN: 0022-1287.
PUB. COUNTRY: ENGLAND: United Kingdom
Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
OTHER SOURCE: GENBANK-M84642; GENBANK-M85087, GENBANK-M85088; GENBANK-M85089; GENBANK-M85090, GENBANK-M85091; GENBANK-M85092; GENBANK-M85093; GENBANK-M85094; GENBANK-M88253
ENTRY MONTH: 199211
ENTRY DATE: Entered STN: 19930122
Last Updated on STN: 19970203
Entered Medline: 19921125

AB The rfb (O antigen) gene cluster of a group C1 Salmonella enterica strain was sequenced; it comprised seven open reading frames which precisely replaced the 16 open reading frames of a group B strain. Two genes of the **mannose** biosynthetic pathway were present: **rfbK** (phosphomannomutase) had a G+C content of 0.61 and had only 40% identity to **rfbK** of group B but was very similar to cpsG of the capsular polysaccharide pathway with 96% identity, whereas **rfbM** [guanosine diphosphomannose (GDP-Man) pyrophosphorylase] had a G+C content of 0.39. Other genes had G+C contents ranging from 0.24 to 0.28. **rfbM**(C1) and **rfbM**(B) had 60% identity, which is much less than expected within a species, but nonetheless indicates a much more recent common ancestor than for **rfbK**. The other genes showed much lower or no similarity to rfb genes of other S. enterica strains. It appears that the gene cluster evolved outside of Salmonella in a species with low G+C content: the **rfbM** gene presumably derives from that period whereas the **rfbK** gene appears to have arisen after transfer of the cluster to S. enterica by duplication of the S. enterica cpsG gene, presumably replacing an **rfbK** gene of low G+C content.

L16 ANSWER 13 OF 16 MEDLINE DUPLICATE 10
ACCESSION NUMBER: 92349966 MEDLINE
DOCUMENT NUMBER: 92349966 PubMed ID: 1379320
TITLE: Molecular analysis of the rfb gene cluster of Salmonella serovar muenchen (strain M67): the genetic basis of the polymorphism between groups C2 and B.
AUTHOR: Brown P K; Romana L K; Reeves P R
CORPORATE SOURCE: Department of Microbiology, University of Sydney, New South Wales, Australia.
SOURCE: MOLECULAR MICROBIOLOGY, (1992 May) 6 (10) 1385-94.
Journal code: MOM; 8712028. ISSN: 0950-382X
PUB. COUNTRY: ENGLAND: United Kingdom
Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
OTHER SOURCE: GENBANK-X61917
ENTRY MONTH: 199208
ENTRY DATE: Entered STN: 19920911
Last Updated on STN: 19980206
Entered Medline: 19920831

AB The rfb (O antigen) gene cluster of group C2 Salmonella differs from that of group B in a central region of 12.4 kb: we report the sequencing of this region of strain M67 (group C2) and a subsequent comparison with the central region of strain LT2 (group B). We find a block of seven open reading frames unique to group C2 which encode the O antigen polymerase (rfc) and the transferases responsible for assembly of the group C2 O antigen. The remaining rfb genes are common to strains M67 and LT2, but rfbJ (CDP-abequose synthase) and **rfbM** and **rfbK** (GDP-**mannose** synthesis), which are immediately adjacent to the central region, are highly divergent. All these genes have a low G+C content and appear to have been recent additions to Salmonella enterica. We discuss the evolutionary significance of the arrangement and divergence of the genes in the polymorphism of the rfb cluster.

L16 ANSWER 14 OF 16 MEDLINE DUPLICATE 11
ACCESSION NUMBER: 92226693 MEDLINE
DOCUMENT NUMBER: 92226693 PubMed ID: 1373435
TITLE: Cloning and structure of group C1 O antigen (rfb gene cluster) from Salmonella enterica serovar montevideo.
AUTHOR: Lee S J; Romana L K; Reeves P R
CORPORATE SOURCE: Department of Microbiology, University of Sydney, NSW, Australia.
SOURCE: JOURNAL OF GENERAL MICROBIOLOGY, (1992 Feb) 138 (Pt 2) 305-12.
Journal code: I87; 0375371. ISSN: 0022-1287
PUB. COUNTRY: ENGLAND: United Kingdom
Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals

ENTRY MONTH: 199205
ENTRY DATE: Entered STN: 19920607
Last Updated on STN: 19970203
Entered Medline: 19920519

AB The Salmonella enterica group C1 O antigen structure has a Man-Man-Man-Man-GlcNAc backbone with a glucose branch, which differs from the S. enterica group B O antigen structure which has a Man-Rha-Gal backbone with abequose as side-chain. We have cloned the group C1 rfb (O antigen) gene cluster from serovar montevideo strain M40, using a low-copy-number cosmid vector. The restriction map of the group C1 (M40) rfb gene cluster was compared with that of group B strain LT2 by Southern hybridization and restriction enzyme analysis. The results indicate that the flanking genes are very similar in the two strains, but there is no detectable similarity in the rfb regions. We localized the **mannose** pathway genes **rfbM** and **rfbK** and one of the genes, **rfbK**, shows considerably similarity to cpsG of strain LT2, suggesting that part of the **mannose** pathway in the group C1 rfb cluster is derived from a gene of the M antigen (cps) cluster. The M antigen, which forms a capsule, is comprised of four sugars, including fucose. The biosynthetic pathway of GDP-fucose has steps in common with the GDP-**mannose** pathway, and the cps cluster has isogenes of **rfbK** and **rfbM**, presumably as part of a fucose pathway. We discuss the structure and possible evolution of the group C1 rfb gene cluster.

L16 ANSWER 15 OF 16 CAPLUS COPYRIGHT 2001 ACS
ACCESSION NUMBER: 1992:421239 CAPLUS
DOCUMENT NUMBER: 117:21239
TITLE: Structure and sequence of the rfb (O antigen) gene cluster of Salmonella serovar typhimurium (strain LT2)
AUTHOR(S): Jiang, X. M.; Neal, B ; Santiago, F.; Lee, S. J.; Romana, L. K., Reeves, P. R.
CORPORATE SOURCE: Dep. Microbiol., Univ Sydney, Sydney, 2006, Australia
SOURCE: Mol. Microbiol. (1991), 5(3), 695-713
CODEN: MOMIEE, ISSN: 0950-382X
DOCUMENT TYPE: Journal
LANGUAGE: English

AB The rfb gene cluster of Salmonella LT2 was cloned and sequenced. The genes rfbA, rfbB, rfbD, rfbF, rfbG, **rfbK**, **rfbM**, rfbP were located individually, and the gene rfbL was located outside the cluster. Approx. 16 open reading frames were found in the region which is essential for the expression of O antigen. The gene products of rfbB and rfbG were homologous with the group of dehydrogenase and related enzymes described previously. Anal. of the G+C ratio of the rfb cluster extended the area of low-G+C compn. previously found in the sequence of rfbJ to the whole rfb gene cluster. Three to five segments with discrete G+C contents and codon adaptation indexes are present in the rfb region, indicating a heterogeneous origin of these segments. Potential promoters were found near the start of the rfb region, supporting the possibility that the rfb gene cluster is an operon.

L16 ANSWER 16 OF 16 MEDLINE DUPLICATE 12
ACCESSION NUMBER: 91287694 MEDLINE
DOCUMENT NUMBER: 91287694 PubMed ID: 1712067
TITLE: The cps gene cluster of Salmonella strain LT2 includes a second **mannose** pathway: sequence of two genes and relationship to genes in the rfb gene cluster.
AUTHOR: Stevenson G; Lee S J; Romana L K; Reeves P R
CORPORATE SOURCE: Department of Microbiology, University of Sydney, N.S.W., Australia.
SOURCE: MOLECULAR AND GENERAL GENETICS, (1991 Jun) 227 (2) 173-80.
Journal code: NGP; 0125036 ISSN: 0026-8925.
PUB. COUNTRY: GERMANY: Germany, Federal Republic of
Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
OTHER SOURCE: GENBANK-X54103; GENBANK-X59886; GENBANK-X63980; GENBANK-X63981; GENBANK-X63982; GENBANK-X63983; GENBANK-X63984; GENBANK-X63985; GENBANK-X63986; GENBANK-X63987
ENTRY MONTH: 199108
ENTRY DATE: Entered STN: 19910825
Last Updated on STN: 19970203
Entered Medline: 19910806

AB We report the presence in Salmonella enterica strain LT2 (serovar thyphimurium) of duplicate genes for two steps in the synthesis of GDP-**mannose**. The previously known genes, **rfbK** (phosphomannomutase) and **rfbM** (**mannose**-1-phosphate guanyltransferase), are part of the gene cluster for the O antigen. The two new genes, cpsB and cpsG, respectively, are thought to be part of the gene cluster for the M antigen capsular polysaccharide, present in many Enterobacteriaceae. The two genes have been sequenced and have a GC

content of 0.61, suggesting an origin outside of Salmonella. Comparison of the inferred protein sequences for cpsB and **rfbM** shows 57% identity of amino acids whereas for cpsG and **rfbK** there is only 19% identity. It is suggested that the greater divergence between cpsG and **rfbK** may be due to a period of accelerated evolution, perhaps precipitated by transfer of the genes from another species.